We claim:

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- A method for the targeted transgenic expression of nucleic
 acid sequences in nonreproductive floral tissues of plants, comprising the following steps,
 - I. introduction of a transgenic expression cassette into plant cells, wherein the transgenic expression cassette comprises at least the following elements
 - a) at least one promoter sequence selected from the group of sequences consisting of
 - i) the promoter sequences of SEQ ID NO: 1 or 2 and
 - ii) functional equivalents of the promoter sequences of SEQ ID NO: 1 or 2 with essentially the same promoter activity as a promoter of SEQ ID NO: 1 or 2 and
 - iii) functional equivalent fragments of the sequences of i) or ii) with essentially the same promoter activity as a promoter of SEQ ID NO: 1 or 2,

and

- b) at least one further nucleic acid sequence, and
- 30 c) optionally further genetic control elements,

wherein at least one promoter sequence and one further nucleic acid sequence are functionally linked together, and the further nucleic acid sequence is heterologous in relation to the promoter sequence, and

- II. selection of transgenic cells which comprise said expression cassette stably integrated into the genome, and
- III. regeneration of complete plants from said transgenic cells, wherein at least one of the further nucleic acid sequences is expressed essentially in all nonreproductive floral tissues, but essentially not in the pollen and the ovaries.

- 2. The method according to claim 1, wherein the functionally equivalent fragment comprises a sequence as shown in SEQ ID NO: 3 or 4.
- 5 3. A method for identifying and/or isolating promoters of genes which encode a promoter having specificity for nonreproductive floral tissue, wherein at least one nucleic acid sequence or a part thereof is employed in the identification and/or isolation, wherein said nucleic acid sequence encodes an amino acid sequences which comprises at least one sequence of SEQ ID NO: 23, 24, 25, 26, 27, 28, 29, 30, 31 or 32 or a variation of these sequences.
- 4. The method according to claim 3, wherein said nucleic acid sequence comprises a sequence of SEQ ID NO: 11, 13, 15, 17, 19 or 21.
- The method according to either of claims 3 or 4, wherein the method is carried out with use of the polymerase chain reaction, and said nucleic acid sequence or a part thereof is employed as primer.
- 6. A method for producing a transgenic expression cassette having specificity for nonreproductive floral tissue,
 25 comprising the following steps:
- I. isolation of a promoter with specificity for nonreproductive floral tissue, where at least one nucleic acid sequence or a part thereof is employed in the isolation, where said nucleic acid sequence encodes an amino acid sequence which comprises at least one sequence as shown in SEQ ID NO: 23, 24, 25, 26, 27, 28, 29, 30, 31 or 32 or a variation of these sequences.
- II. functional linkage of said promoter with a further nucleic acid sequence, where said nucleic acid sequence is heterologous in relation to the promoter.
- 7. The method according to claim 6, where said nucleic acid sequence comprises a sequence as shown in SEQ ID NO: 11, 13, 15, 17, 19 or 21.
- 8. The method according to either of claims 6 or 7, where the method is carried out with use of the polymerase chain reaction, and said nucleic acid sequence or a part thereof is employed as primer.

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- 9. A polypeptide comprising an amino acid sequence of SEQ ID NO: 16, 18, 20 or 22.
- 10. A nucleic acid sequence encoding a polypeptide according to claim 9.
 - 11. The nucleic acid sequence according to claim 10, comprising a sequence selected from the group of sequences of SEQ ID NO: 15, 17, 19 or 21 and the sequences derived therefrom as the result of the degeneracy of the genetic code.
- 12. The use of at least one nucleic acid sequence or a part thereof in methods for identifying and/or isolating promoters with specificity for the nonreproductive floral tissue, where said nucleic acid sequence encodes an amino acid sequence comprising at least one sequence of SEQ ID NO: 23, 24, 25, 26, 27, 28, 29, 30, 31 or 32 or a variation of these sequences.
- 20 13. The use according to claim 12, where said nucleic acid sequence comprises a sequence of SEQ ID NO: 11, 13, 15, 17, 19 or 21.
- 14. A transgenic expression cassette for the targeted transgenic
 25 expression of nucleic acid sequences in nonreproductive floral tissues of plants, comprising
 - a) at least one promoter sequence selected from the group of sequences consisting of
 - i) the promoter sequences of SEQ ID NO: 1 or 2 and
 - ii) functional equivalents of the promoter sequences of SEQ ID NO: 1 or 2 with essentially the same promoter activity as a promoter of SEQ ID NO: 1 or 2 and
 - iii) functionally equivalent fragments of the sequences ofi) or ii) with essentially the same promoter activityas a promoter of SEQ ID NO: 1 or 2,

and

- b) at least one further nucleic acid sequence, and
- c) optionally further genetic control elements,

where at least one promoter sequence and one further nucleic acid sequence are functionally linked together, and the further nucleic acid sequence is heterologous in relation to the promoter sequence.

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- 15. The transgenic expression cassette according to claim 14, wherein the functionally equivalent fragment comprises a sequence of SEQ ID NO: 3 or 4.
- 10 16. The transgenic expression cassette according to claim 14 or 15, where
 - a) the nucleic acid sequence to be expressed is functionally linked with further genetic control sequences, or

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- b) the expression cassette comprises additional functional elements, or
- c) a) and b) apply.

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- 17. The transgenic expression cassette according to one of claims 14 to 16, wherein the nucleic acid sequence to be expressed transgenically makes possible
- 25 a) the expression of a protein encoded by said nucleic acid sequence, or
- b) the expression of a sense-RNA, anti-sense-RNA or double-stranded RNA encoded by said nucleic acid
 30 sequence.
 - 18. The transgenic expression cassette according to one of claims 14 to 17, wherein the nucleic acid sequence to be expressed transgenically is selected from the group of nucleic acid
- sequences encoding chalcone synthases, phenylalanine ammonium lyases, photolyases, deoxyxylulose-5-phosphate synthases, phytoene synthases, phytoene desaturases, lycopene cyclases, hydroxylases, "antifreeze" polypeptides, CBF1-transcription activators, glutamate dehydrogenases, calcium-dependent
- protein kinases, calcineurin, farnesyltransferases, ferritin, oxalate oxidases, DREBLA factor, trehalose-phosphate phosphatases, chitinases, glucanases, ribosome-inactivating protein, lysozyme, Bacillus thuringiensis endotoxins, amylase inhibitors, protease inhibitors, lectins, RNAses, ribozymes,
- endochitinase, cytochrome P-450, acetyl-CoA carboxylases, amino acid transporters, monosaccharide-transporters, lycopine cyklases, carotene ketolases, endoxyloglucan

transferases, $\Delta 6$ -acyllipid desaturases, $\Delta 6$ -desaturases, $\Delta 5$ -fatty acid desaturases, $\Delta 6$ -elongases and IPP-isomerases.

19. The transgenic expression cassette according to one of claims 14 to 18, wherein the nucleic acid sequence to be expressed transgenically is selected from the group of nucleic acid sequences described by GenBank Acc.-No.: M20308, BAB00748, U62549, U77378, S78423, U32624, L25042, X92657, AJ002399, D45881, AF163819, AB044391, AJ222980 and AF078796.

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- 20. A transgenic expression vector comprising an expression cassette according to one of claims 14 to 19.
- 21. A transgenic organism, transformed with an expression15 cassette of claims 14 to 19 or an expression vector of claim 20.
- 22. The transgenic organism according to claim 21 selected from the group consisting of bacteria, yeasts, fungi, non-human animal and plant organisms or of cells, cell cultures, parts, tissues, organs or propagation material derived therefrom.
 - 23. The transgenic organism as claimed in claim 21 or 22 selected from the group of agricultural crop plants.

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24. The use of a transgenic organism according to any of claims. 21 to 23 or cells, cell cultures, parts, tissues, organs or propagation material derived therefrom to produce human or animal foods, seeds, pharmaceuticals or fine chemicals.

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25. A method for producing pharmaceuticals or fine chemicals in transgenic organisms according to one of claims 21 to 23 or cells, cell cultures, parts, tissues, organs or propagation material derived therefrom, where the transgenic organism or cells, cell cultures, parts, tissues, organs or propagation material derived from them is/are cultured or grown, and the desired pharmaceutical or the desired fine chemical is isolated.

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Transgenic expression cassettes for expressing nucleic acids in nonreproductive floral tissues of plants

Abstract

The invention relates to methods for the targeted transgenic expression of nucleic acid sequences in nonreproductive floral tissues of plants, and to transgenic expression cassettes and expression vectors which comprise promoters having an expression specificity for nonreproductive tissues of the flower. The invention further relates to organisms (preferably plants) transformed with these transgenic expression cassettes or expression vectors, to cultures, parts or propagation material derived therefrom, and to the use of the same for producing human and animal foods, seeds, pharmaceuticals or fine chemicals.